APPLICATION

FOR

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REINFORCED FOAM IMPLANTS WITH ENHANCED INTEGRITY FOR SOFT TISSUE REPAIR AND REGENERATION

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REINFORCED FOAM IMPLANTS WITH ENHANCED INTEGRITY FOR SOFT TISSUE REPAIR AND REGENERATION

FIELD OF THE INVENTION

The present invention relates to bioabsorbable, porous, reinforced, biocompatible tissue repair stimulating implant devices comprising at least one biological component for use in the repair of orthopaedic type injuries, such as damage to the meniscus and rotator cuff, and methods for making such devices.

BACKGROUND OF THE INVENTION

Individuals can sometimes sustain an injury to tissue, such as cartilage, muscle, bone, and sinew that requires repair by surgical intervention. Such repairs can be effected by suturing or otherwise repairing the damaged tissue, and/or by augmenting the damaged tissue with other tissue or with a tissue implant. The implant can provide structural support to the damaged tissue.

One example of a common tissue injury concerns damage to cartilage, for example, the menisci of a knee joint. There are two menisci of the knee joint, a medial and a lateral meniscus. The meniscus is a biconcave, fibrocartilage tissue that is interposed between the femur and tibia of the leg. The primary functions of the meniscus are to bear loads, absorb shock, stabilize, and lubricate the joint. If not treated properly, an injury to the meniscus, such as a "bucket-handle tear," can lead to the development of osteoarthritis. Currently, treatment modalities for a damaged meniscus include removal of the meniscus and surgical repair of the damaged meniscus.

Another common tissue injury is a damaged or torn rotator cuff, which facilitates circular motion of the humerus bone relative to the scapula. The most common injury associated with the rotator cuff is a strain or tear to the supraspinatus tendon. This tear can be at the insertion site of the tendon with the humerus, thereby releasing the tendon partially, or fully (depending upon the severity of the injury), from the bone. Additionally, the strain or tear can occur within the tendon itself. Treatment for a strained tendon usually involves physical cessation from use of the tendon. However, depending upon the severity of the injury, a torn tendon might require surgical intervention as in the case of a full tear of the supraspinatus tendon from the humerus. Surgical intervention can involve the repair and/or reattachment of torn tissue. A prolonged recovery period often follows repair of a rotator cuff tear.

Surgical treatment of damaged tissue (e.g., the menisci or rotator cuff) would benefit from techniques that effect a more reliable repair of tissue, and which facilitate more rapid healing. Thus, various implants have been used in surgical procedures to help achieve these benefits. Examples of such implants include those that are made from biologically derived tissue (e.g., allografts and autografts), and those that are synthetic. Biologically derived materials can have disadvantages in that they can contribute to disease transmission, while synthetic materials are difficult to manufacture in such a way that their properties are reproducible from batch to batch.

Various known devices and techniques for treating such conditions have been described in the prior art. For example, Naughton et al. (U.S. Pat. 5,842,477) describe an *in vivo* method of making and/or repairing cartilage by implanting a biocompatible structure in combination with periosteal/perichondrial tissue which facilitates the securing of the implant.

Various tissue reinforcing materials are disclosed in U.S. Patent No. 5,891,558 (Bell et al.) and European Patent Application No. 0 274 898 A2 (Hinsch). Bell et al. describe biopolymer foams and foam constructs that can be used in tissue repair and reconstruction. Hinsch describes an open cell, foam-like implant made from resorbable materials, which has one or more textile reinforcing elements embedded therein. Although potentially useful, the implant material is believed to lack sufficient strength and structural integrity to be effectively used as a tissue repair implant.

Despite existing technology, there continues to be a need for devices and methods for securing damaged tissue and facilitating rapid healing of the damaged tissue.

SUMMARY OF THE INVENTION

This invention relates to bioabsorbable, porous, reinforced, biocompatible tissue repair stimulating implant, or "scaffold," devices for use in the repair and/or regeneration of diseased or damaged tissue, and the methods for making and using these devices. The implants comprise a bioabsorable polymeric foam component having pores with an open cell pore structure. The foam component is reinforced with a material such as a mesh. Preferably, the implant has sufficient structural integrity to enable it to be handled in the operating room prior to and during implantation. These implants should also have sufficient properties (e.g., tear strength) to enable them to accept and retain sutures or other fasteners without tearing. Desirable properties are imparted to the implant of the invention by integrating the foam component with the reinforcement component. That is, the pore-forming webs or walls of the foam component penetrate the mesh of the reinforcement component so as to interlock therewith. The implant may include one or more layers of each of the foam and

reinforcement components. Preferably, adjacent layers of foam are also integrated by at least a partial interlocking of the pore-forming webs or walls in the adjacant layers. The implants of the instant invention further comprise at least one biological component that is incorporated therein.

The reinforcement material is preferably a mesh, which may be bioabsorbable. The reinforcement should have a sufficient mesh density to permit suturing, but the density should not be so great as to impede proper bonding between the foam and the reinforcement. A preferred mesh density is in the range of about 12 to 80%.

The biological component of the present invention comprises at least one effector molecule and/or cell, which contributes to the healing process of an injured tissue. Collectively, these materials are sometimes referred to herein as "effectors." The effectors can be a cellular factor such as a protein or peptide (for the sake of simplicity, use of the term "protein" herein will include peptide), a non-protein biomolecule, a cell type, a pharmaceutical agent, or combinations thereof. One function of the implant of the current invention is as a carrier for the effectors, and the effector can be incorporated within the implant either prior to or following surgical placement of the implant.

The invention also relates to a method of preparing such biocompatible, bioabsorbable tissue repair stimulating implants. The implants are made by placing a reinforcement material within a mold in a desired position and orientation. A solution of a desired polymeric material in a suitable solvent is added to the mold and the solution is lyophilized to obtain the implant in which a reinforcement material is embedded in a polymeric foam. The effector may be added to the implant, either during or after manufacture, by a variety of techniques.

The tissue repair stimulating implant can be used to treat injuries ocurring within the musculoskeletal system, such as injuries to the rotator cuff or meniscus.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention will be more fully understood by reference to the following detailed description when considered in conjunction with the accompanying drawings, in which:

Figure 1 is a sectional view of a tissue implant constructed according to the present invention;

Figure 2 is a sectional view of an alternative embodiment of the implant of the present invention;

Figure 3 is a sectional view of yet another embodiment of the implant of the present invention;

Figure 4 is a perspective view of one embodiment of a mold set-up useful with the present invention;

Figure 5 is a sectional view of a portion of the mold set-up of Figure 4;

Figure 6 is a scanning electron micrograph of a bioabsorbable knitted mesh reinforcement material useful with the implant of the present invention; and

Figure 7 is a scanning electron micrograph of a portion of an implant according to the present invention.

DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to a biocompatible tissue repair stimulating implant or "scaffold" device which, preferably, is bioabsorbable, and to methods for making and using

such a device. The implant includes one or more layers of a bioabsorbable polymeric foam having pores with an open cell pore structure. A reinforcement component is also present within the implant to contribute enhanced mechanical and handling properties. The reinforcement component is preferably in the form of a mesh fabric that is biocompatible. The reinforcement component may be bioabsorbable as well. The implant has incorporated therein a biological component, or effector that assists in and/or expedites tissue healing. Preferably, the biological component is housed primarily within the pores of the foam component of the implant.

In some surgical applications, such as for use in the repair of a torn rotator cuff or meniscus, the tissue implants of the invention must be able to be handled in the operating room, and they must be able to be sutured or otherwise fastened without tearing. Additionally, the implants should have a burst strength adequate to reinforce the tissue, and the structure of the implant must be suitable to encourage tissue ingrowth. A preferred tissue ingrowth-promoting structure is one where the cells of the foam component are open and sufficiently sized to permit cell ingrowth and to house the effector. A suitable pore size to accommodate these features is one in which the pores have an average diameter in the range of about 100 to 1000 microns and, more preferably, about 150 to 500 microns.

Referring to FIGS. 1 through 3, the implant 10 includes a polymeric foam component 12 and a reinforcement component 14. The foam component preferably has pores 13 with an open cell pore structure. Although illustrated as having the reinforcement component disposed substantially in the center of a cross section of the implant, it is understood that the reinforcement material can be disposed at any location within the implant. Further, as shown in FIG. 2, more than one layer of each of the foam component 12a, 12b and reinforcement component 14a, 14b may be present in the implant. It is understood that various layers of the

foam component and/or the reinforcement materials may be made from different materials and have different pore sizes.

FIG. 3 illustrates an embodiment in which a barrier layer 16 is present in the implant. Although illustrated as being only on one surface of the implant 10, the barrier layer 16 may be present on either or both of the top and bottom surfaces 18, 20 of the implant.

The implant 10 must have sufficient structural integrity and physical properties to facilitate ease of handling in an operating room environment, and to permit it to accept and retain sutures or other fasteners without tearing. Adequate strength and physical properties are developed in the implant through the selection of materials used to form the foam and reinforcement components, and the manufacturing process. As shown in FIG. 7, the foam component 12 is integrated with the reinforcement component 14 such that the web or walls of the foam componenets that form pores 13 penetrate the mesh of the reinforcement component 14 and interlock with the reinforcement component. The pore-forming walls in adjacent layers of the foam component also interlock with one another, regardless of whether the foam layers are separated by a layer of reinforcement materials or whether they are made of the same or different materials.

A variety of bioabsorbable polymers can be used to make porous, reinforced tissue repair stimulating implant or scaffold devices according to the present invention. Examples of suitable biocompatible, bioabsorbable polymers include polymers selected from the group consisting of aliphatic polyesters, poly(amino acids), copoly(ether-esters), polyalkylenes oxalates, polyamides, tyrosine derived polycarbonates, poly(iminocarbonates), polyorthoesters, polyoxaesters, polyamidoesters, polyoxaesters containing amine groups, poly(anhydrides), polyphosphazenes, biomolecules (i.e., biopolymers such as collagen, elastin,

bioabsorbable starches, etc.) and blends thereof. For the purpose of this invention aliphatic polyesters include, but are not limited to, homopolymers and copolymers of lactide (which includes lactic acid, D-,L- and meso lactide), glycolide (including glycolic acid), ε-caprolactone, p-dioxanone (1,4-dioxan-2-one), trimethylene carbonate (1,3-dioxan-2-one), alkyl derivatives of trimethylene carbonate, δ-valerolactone, β-butyrolactone, γ-butyrolactone, ε-decalactone, hydroxybutyrate, hydroxyvalerate, 1,4-dioxepan-2-one (including its dimer 1,5,8,12-tetraoxacyclotetradecane-7,14-dione), 1,5-dioxepan-2-one, 6,6-dimethyl-1,4-dioxan-2-one 2,5-diketomorpholine, pivalolactone, α, α-diethylpropiolactone, ethylene carbonate, ethylene oxalate, 3-methyl-1,4-dioxane-2,5-dione, 3,3-diethyl-1,4-dioxan-2,5-dione, 6,8-dioxabicycloctane-7-one and polymer blends thereof. Poly(iminocarbonates), for the purpose of this invention, are understood to include those polymers as described by Kemnitzer and Kohn, in the Handbook of Biodegradable Polymers, edited by Domb, et. al., Hardwood Academic Press, pp. 251-272 (1997). Copoly(ether-esters), for the purpose of this invention, are understood to include those copolyester-ethers as described in the Journal of Biomaterials Research, Vol. 22, pages 993-1009, 1988 by Cohn and Younes, and in Polymer Preprints (ACS Division of Polymer Chemistry), Vol. 30(1), page 498, 1989 by Cohn (e.g., PEO/PLA). Polyalkylene oxalates, for the purpose of this invention, include those described in U.S. Patent Numbers 4,208,511; 4,141,087; 4,130,639; 4,140,678; 4,105,034; and 4,205,399. Polyphosphazenes, co-, ter- and higher order mixed monomer based polymers made from L-lactide, D,L-lactide, lactic acid, glycolide, glycolic acid, para-dioxanone, trimethylene carbonate and ϵ-caprolactone such as are described by Allcock in The Encyclopedia of Polymer Science, Vol. 13, pages 31-41, Wiley Intersciences, John Wiley & Sons, 1988 and by Vandorpe, et al in the Handbook of Biodegradable Polymers, edited by Domb, et al., Hardwood Academic Press, pp. 161-182 (1997). Polyanhydrides include those derived from diacids of the form HOOC-C_6H_4 –O-$(CH_2)_m$-O-C_6H_4-COOH, where "m" is an integer in the range of from 2 to 8, and copolymers thereof with aliphatic alpha-omega diacids

of up to 12 carbons. Polyoxaesters, polyoxaamides and polyoxaesters containing amines and/or amido groups are described in one or more of the following U.S. Patent Nos. 5,464,929; 5,595,751; 5,597,579; 5,607,687; 5,618,552; 5,620,698; 5,645,850; 5,648,088; 5,698,213; 5,700,583; and 5,859,150. Polyorthoesters such as those described by Heller in Handbook of Biodegradable Polymers, edited by Domb, et al., Hardwood Academic Press, pp. 99-118 (1997).

As used herein, the term "glycolide" is understood to include polyglycolic acid. Further, the term "lactide" is understood to include L-lactide, D-lactide, blends thereof, and lactic acid polymers and copolymers.

Currently, aliphatic polyesters are among the preferred absorbable polymers for use in making the foam implants according to the present invention. Aliphatic polyesters can be homopolymers, copolymers (random, block, segmented, tapered blocks, graft, triblock, etc.) having a linear, branched or star structure. Suitable monomers for making aliphatic homopolymers and copolymers may be selected from the group consisting of, but are not limited, to lactic acid, lactide (including L-, D-, meso and D,L mixtures), glycolic acid, glycolide, ε-caprolactone, p-dioxanone (1,4-dioxan-2-one), trimethylene carbonate (1,3-dioxan-2-one), δ-valerolactone, β-butyrolactone, ϵ-decalactone, 2,5-diketomorpholine, pivalolactone, α, α-diethylpropiolactone, ethylene carbonate, ethylene oxalate, 3-methyl-1,4-dioxane-2,5-dione, 3,3-diethyl-1,4-dioxan-2,5-dione, γ-butyrolactone, 1,4-dioxepan-2-one, 1,5-dioxepan-2-one, 6,6-dimethyl-dioxepan-2-one, 6,8-dioxabicycloctane-7-one, and combinations thereof.

Elastomeric copolymers are also particularly useful in the present invention. Suitable elastomeric polymers include those with an inherent viscosity in the range of about 1.2 dL/g to

4 dL/g, more preferably about 1.2 dL/g to 2 dL/g and most preferably about 1.4 dL/g to 2 dL/g as determined at 25°C in a 0.1 gram per deciliter (g/dL) solution of polymer in hexafluoroisopropanol (HFIP). Further, suitable elastomers exhibit a high percent elongation and a low modulus, while possessing good tensile strength and good recovery characteristics. In the preferred embodiments of this invention, the elastomer from which the foam component is formed exhibits a percent elongation (e.g., greater than about 200 percent and preferably greater than about 500 percent). In addition to these elongation and modulus properties, suitable elastomers should also have a tensile strength greater than about 500 psi, preferably greater than about 1,000 psi, and a tear strength of greater than about 50 lbs/inch, preferably greater than about 80 lbs/inch.

Exemplary bioabsorbable, biocompatible elastomers include, but are not limited to, elastomeric copolymers of ε-caprolactone and glycolide (including polyglycolic acid) with a mole ratio of ε-caprolactone to glycolide of from about 35:65 to about 65:35, more preferably from 45:55 to 35:65; elastomeric copolymers of ε-caprolactone and lactide (including L-lactide, D-lactide, blends thereof, and lactic acid polymers and copolymers) where the mole ratio of ε-caprolactone to lactide is from about 35:65 to about 65:35 and more preferably from 45:55 to 30:70 or from about 95:5 to about 85:15; elastomeric copolymers of p-dioxanone (1,4-dioxan-2-one) and lactide (including L-lactide, D-lactide, blends thereof, and lactic acid polymers and copolymers) where the mole ratio of p-dioxanone to lactide is from about 40:60 to about 60:40; elastomeric copolymers of ε-caprolactone and p-dioxanone where the mole ratio of ϵ-caprolactone to p-dioxanone is from about from 30:70 to about 70:30; elastomeric copolymers of p-dioxanone and trimethylene carbonate where the mole ratio of p-dioxanone to trimethylene carbonate is from about 30:70 to about 70:30; elastomeric copolymers of trimethylene carbonate and glycolide (including polyglycolic acid) where the mole ratio of trimethylene carbonate to glycolide is from about 30:70 to about 70:30; elastomeric

copolymers of trimethylene carbonate and lactide (including L-lactide, D-lactide, blends thereof, and lactic acid polymers and copolymers) where the mole ratio of trimethylene carbonate to lactide is from about 30:70 to about 70:30; and blends thereof. Examples of suitable bioabsorbable elastomers are described in U.S. Patent Nos. 4,045,418; 4,057,537 and 5,468,253.

In one embodiment, the elastomer is a 35:65 copolymer of polyglycolic acid and polycaprolactone, formed in a dioxane solvent and including a polydioxanone mesh. In another embodiment, the elastomer is a 50:50 blend of a 35:65 copolymer of polyglycolic acid and polycaprolactone and 40:60 ϵ-caprolactone-co-lactide.

One of ordinary skill in the art will appreciate that the selection of a suitable polymer or copolymer for forming the foam depends on several factors. The more relevant factors in the selection of the appropriate polymer(s) that is used to form the foam component include bioabsorption (or bio-degradation) kinetics; *in vivo* mechanical performance; cell response to the material in terms of cell attachment, proliferation, migration and differentiation; and biocompatibility. Other relevant factors, which to some extent dictate the *in vitro* and *in vivo* behavior of the polymer, include the chemical composition, spatial distribution of the constituents, the molecular weight of the polymer, and the degree of crystallinity.

The ability of the material substrate to resorb in a timely fashion in the body environment is critical. But the differences in the absorption time under *in vivo* conditions can also be the basis for combining two different copolymers. For example, a copolymer of 35:65 ε-caprolactone and glycolide (a relatively fast absorbing polymer) is blended with 40:60 ε-caprolactone and L-lactide copolymer (a relatively slow absorbing polymer) to form a foam component. Depending upon the processing technique used, the two constituents can be either

randomly inter-connected bicontinuous phases, or the constituents could have a gradient-like architecture in the form of a laminate type composite with a well integrated interface between the two constituent layers. The microstructure of these foams can be optimized to regenerate or repair the desired anatomical features of the tissue that is being engineered.

In one embodiment, it is desirable to use polymer blends to form structures which transition from one composition to another composition in a gradient-like architecture. Foams having this gradient-like architecture are particularly advantageous in tissue engineering applications to repair or regenerate the structure of naturally occurring tissue such as cartilage (articular, meniscal, septal, tracheal, etc.), rotator cuff, esophagus, skin, bone, and vascular tissue. For example, by blending an elastomer of ε-caprolactone-co-glycolide with ε-caprolactone-co-lactide (e.g., with a mole ratio of about 5:95) a foam may be formed that transitions from a softer spongy material to a stiffer more rigid material in a manner similar to the transition from cartilage to bone. Clearly, one of ordinary skill in the art will appreciate that other polymer blends may be used for similar gradient effects, or to provide different gradients (e.g., different absorption profiles, stress response profiles, or different degrees of elasticity). For example, such design features can establish a concentration gradient for the biological component or effector such that a higher concentration of the effector is present in one region of the implant (e.g., an interior portion) than in another region (e.g., outer portions). This may be effected by engineering an implant in which the overall pore volume is greater in a region in which it is desired to have a greater concentration of biological component.

The implants of the invention can also be used for organ repair replacement or regeneration strategies that may benefit from these unique tissue implants. For example, these implants can be used for spinal disc, cranial tissue, dura, nerve tissue, liver, pancreas, kidney,

bladder, spleen, cardiac muscle, skeletal muscle, skin, fascia, maxillofacial, stomach, tendons, cartilage, ligaments, and breast tissues.

The reinforcing component of the tissue repair stimulating implant of the present invention can be comprised of any absorbable or non-absorbable biocompatible material, including textiles with woven, knitted, warped knitted (i.e., lace-like), non-woven, and braided structures. In an exemplary embodiment, the reinforcing component has a mesh-like structure. In any of the above structures, mechanical properties of the material can be altered by changing the density or texture of the material, or by embedding particles in the material. The fibers used to make the reinforcing component can be monofilaments, yarns, threads, braids, or bundles of fibers. These fibers can be made of any biocompatible material including bioabsorbable materials such as polylactic acid (PLA), polyglycolic acid (PGA), polycaprolactone (PCL), polydioxanone (PDO), trimethylene carbonate (TMC), polyvinyl alcohol (PVA), copolymers or blends thereof. In one embodiment, the fibers are formed of a polylactic acid and polyglycolic acid copolymer at a 95:5 mole ratio.

In another embodiment, the fibers that form the reinforcing material can be made of a bioabsorbable glass. Bioglass, a silicate containing calcium phosphate glass, or calcium phosphate glass with varying amounts of solid particles added to control resorption time are examples of materials that could be spun into glass fibers and used for the reinforcing material. Suitable solid particles that may be added include iron, magnesium, sodium, potassium, and combinations thereof.

The reinforcing material may also be formed from a thin, perforation-containing elastomeric sheet with perforations to allow tissue ingrowth. Such a sheet could be made of

blends or copolymers of polylactic acid (PLA), polyglycolic acid (PGA), polycaprolactone (PCL), and polydioxanone (PDO).

In one embodiment, filaments that form the reinforcing material may be co-extruded to produce a filament with a sheath/core construction. Such filaments are comprised of a sheath of biodegradable polymer that surrounds one or more cores comprised of another biodegradable polymer. Filaments with a fast-absorbing sheath surrounding a slower-absorbing core may be desirable in instances where extended support is necessary for tissue ingrowth.

One of ordinary skill in the art will appreciate that one or more layers of the reinforcing material may be used to reinforce the tissue implant of the invention. In addition, biodegradable reinforcing layers (e.g., meshes) of the same structure and chemistry or different structures and chemistries can be overlaid on top of one another to fabricate reinforced tissue implants with superior mechanical strength.

The biological component that is incorporated within the implant can be selected from among a variety of effectors that, when present at the site of injury, promote healing and/or regeneration of the affected tissue. In addition to being compounds or agents that actually promote or expedite healing, the effectors may also include compounds or agents that prevent infection (e.g., antimicrobial agents and antibiotics), compounds or agents that reduce inflammation (e.g., anti-inflammatory agents), and compounds or agents that suppress the immune system (e.g., immunosuppressants). By way of example, other types of effectors present within the implant of the present invention include heterologous or autologous growth factors, proteins, glycoproteins, hormones, cytokines, glycosaminoglycans, nucleic acids,

analgesics, and cell types. It is understood that one or more effectors of the same or different functionality may be incorporated within the implant.

Examples of suitable effectors include the multitude of heterologous or autologous growth factors known to promote healing and/or regeneration of injured or damaged tissue. Exemplary growth factors include, but are not limited to, TGF-β, bone morphogenic protein, fibroblast growth factor, platelet-derived growth factor, vascular endothelial cell-derived growth factor (VEGF), epidermal growth factor, insulin-like growth factor, hepatocyte growth factor, and fragments thereof.

The proteins that may be present within the implant include proteins that are secreted from a cell which is housed within the implant, as well as those that are present within the implant in an isolated form. The isolated form of a protein typically is one that is about 55% or greater in purity, i.e., isolated from other cellular proteins, molecules, debris, etc. More preferably, the isolated protein is one that is at least 65% pure, and most preferably one that is at least about 75 to 95% pure. Notwithstanding the above, one of ordinary skill in the art will appreciate that proteins having a purity below about 55% are still considered to be within the scope of this invention. As used herein, the term "protein" embraces glycoproteins, lipoproteins, proteoglycans, peptides, and fragments thereof. Examples of proteins useful as effectors include, but are not limited to, pleiotrophin, endothelin, tenascin, fibronectin, fibrinogen, vitronectin, V-CAM, I-CAM, N-CAM, selectin, cadherin, integrin, laminin, actin, myosin, collagen, microfilament, intermediate filament, antibody, elastin, fibrillin, and fragments thereof.

Glycosaminoglycans, highly charged polysaccharides which play a role in cellular adhesion, may also serve as effectors according to the present invention. Exemplary

glycosaminoglycans useful as effectors include, but are not limited to, heparan sulfate, heparin, chondroitin sulfate, dermatan sulfate, keratin sulfate, hyaluronan (also known as hyaluronic acid), and combinations thereof.

Suitable cell types that can serve as effectors according to this invention include, but are not limited to, osteocytes, osteoblasts, osteoclasts, fibroblasts, stem cells, pluripotent cells, chondrocyte progenitors, chondrocytes, endothelial cells, macrophages, leukocytes, adipocytes, monocytes, plasma cells, mast cells, umbilical cord cells, stromal cells, mesenchymal stem cells, epithelial cells, myoblasts, and bone marrow cells. Cells typically have at their surface receptor molecules which are responsive to a cognate ligand (e.g., a stimulator). A stimulator is a ligand which when in contact with its cognate receptor induce the cell possessing the receptor to produce a specific biological action. For example, in response to a stimulator (or ligand) a cell may produce significant levels of secondary messengers, like Ca^{+2}, which then will have subsequent effects upon cellular processes such as the phosphorylation of proteins, such as (keeping with our example) protein kinase C. In some instances, once a cell is stimulated with the proper stimulator, the cell secretes a cellular messenger usually in the form of a protein (including glycoproteins, proteoglycans, and lipoproteins). This cellular messenger can be an antibody (e.g., secreted from plasma cells), a hormone, (e.g., a paracrine, autocrine, or exocrine hormone), or a cytokine.

The foam component of the tissue implant may be formed as a foam by a variety of techniques well known to those having ordinary skill in the art. For example, the polymeric starting materials may be foamed by lyophilization, supercritical solvent foaming (i.e., as described in EP 464,163), gas injection extrusion, gas injection molding or casting with an extractable material (e.g., salts, sugar or similar suitable materials).

In one embodiment, the foam component of the engineered tissue repair stimulating implant devices of the present invention may be made by a polymer-solvent phase separation technique, such as lyophilization. Generally, however, a polymer solution can be separated into two phases by any one of the four techniques: (a) thermally induced gelation/ crystallization; (b) non-solvent induced separation of solvent and polymer phases; (c) chemically induced phase separation, and (d) thermally induced spinodal decomposition. The polymer solution is separated in a controlled manner into either two distinct phases or two bicontinuous phases. Subsequent removal of the solvent phase usually leaves a porous structure of density less than the bulk polymer and pores in the micrometer ranges. See Microcellular Foams Via Phase Separation, J. Vac. Sci. Technolol., A. T. Young, Vol. 4(3), May/Jun 1986.

The steps involved in the preparation of these foams include choosing the right solvents for the polymers to be lyophilized and preparing a homogeneous solution. Next, the polymer solution is subjected to a freezing and vacuum drying cycle. The freezing step phase separates the polymer solution and vacuum drying step removes the solvent by sublimation and/or drying, leaving a porous polymer structure or an interconnected open cell porous foam.

Suitable solvents that may be used in the preparation of the foam component include, but are not limited to, formic acid, ethyl formate, acetic acid, hexafluoroisopropanol (HFIP), cyclic ethers (e.g., tetrahydrofuran (THF), dimethylene fluoride (DMF), and polydioxanone (PDO)), acetone, acetates of C2 to C5 alcohols (e.g., ethyl acetate and t-butylacetate), glyme (e.g., monoglyme, ethyl glyme, diglyme, ethyl diglyme, triglyme, butyl diglyme and tetraglyme), methylethyl ketone, dipropyleneglycol methyl ether, lactones (e.g., γ-valerolactone, δ-valerolactone, β-butyrolactone, γ-butyrolactone), 1,4-dioxane, 1,3-dioxolane, 1,3-dioxolane-2-one (ethylene carbonate), dimethlycarbonate, benzene, toluene, benzyl

alcohol, p-xylene, naphthalene, tetrahydrofuran, N-methyl pyrrolidone, dimethylformamide, chloroform, 1,2-dichloromethane, morpholine, dimethylsulfoxide, hexafluoroacetone sesquihydrate (HFAS), anisole and mixtures thereof. Among these solvents, a preferred solvent is 1,4-dioxane. A homogeneous solution of the polymer in the solvent is prepared using standard techniques.

The applicable polymer concentration or amount of solvent that may be utilized will vary with each system. Generally, the amount of polymer in the solution can vary from about 0.5% to about 90% and, preferably, will vary from about 0.5% to about 30% by weight, depending on factors such as the solubility of the polymer in a given solvent and the final properties desired in the foam.

In one embodiment, solids may be added to the polymer-solvent system to modify the composition of the resulting foam surfaces. As the added particles settle out of solution to the bottom surface, regions will be created that will have the composition of the added solids, not the foamed polymeric material. Alternatively, the added solids may be more concentrated in desired regions (i.e., near the top, sides, or bottom) of the resulting tissue implant, thus causing compositional changes in all such regions. For example, concentration of solids in selected locations can be accomplished by adding metallic solids to a solution placed in a mold made of a magnetic material (or vice versa).

A variety of types of solids can be added to the polymer-solvent system. Preferably, the solids are of a type that will not react with the polymer or the solvent. Generally, the added solids have an average diameter of less than about 1.0 mm and preferably will have an average diameter of about 50 to about 500 microns. Preferably, the solids are present in an amount such that they will constitute from about 1 to about 50 volume percent of the total

volume of the particle and polymer-solvent mixture (wherein the total volume percent equals 100 volume percent).

Exemplary solids include, but are not limited to, particles of demineralized bone, calcium phosphate particles, Bioglass particles, calcium sulfate, or calcium carbonate particles for bone repair, leachable solids for pore creation and particles of bioabsorbable polymers not soluble in the solvent system that are effective as reinforcing materials or to create pores as they are absorbed, and non-bioabsorbable materials.

Suitable leachable solids include nontoxic leachable materials such as salts (e.g., sodium chloride, potassium chloride, calcium chloride, sodium tartrate, sodium citrate, and the like), biocompatible mono and disaccharides (e.g., glucose, fructose, dextrose, maltose, lactose and sucrose), polysaccharides (e.g., starch, alginate, chitosan), water soluble proteins (e.g., gelatin and agarose). The leachable materials can be removed by immersing the foam with the leachable material in a solvent in which the particle is soluble for a sufficient amount of time to allow leaching of substantially all of the particles, but which does not dissolve or detrimentally alter the foam. The preferred extraction solvent is water, most preferably distilled-deionized water. Such a process is described in U.S. Patent No. 5,514,378. Preferably the foam will be dried after the leaching process is complete at low temperature and/or vacuum to minimize hydrolysis of the foam unless accelerated absorption of the foam is desired.

Suitable non-bioabsorbable materials include biocompatible metals such as stainless steel, cobalt chrome, titanium and titanium alloys, and bioinert ceramic particles (e.g., alumina, zirconia, and calcium sulfate particles). Further, the non-bioabsorbable materials may include polymers such as polyethylene, polyvinylacetate, polymethylmethacrylate,

silicone, polyethylene oxide, polyethylene glycol, polyurethanes, polyvinyl alcohol, natural biopolymers (e.g., cellulose particles, chitin, keratin, silk, and collagen particles), and fluorinated polymers and copolymers (e.g., polyvinylidene fluoride, polytetrafluoroethylene, and hexafluoropropylene).

It is also possible to add solids (e.g., barium sulfate) that will render the tissue implants radio opaque. The solids that may be added also include those that will promote tissue regeneration or regrowth, as well as those that act as buffers, reinforcing materials or porosity modifiers.

As noted above, porous, reinforced tissue repair stimulating implant devices of the present invention are made by injecting, pouring, or otherwise placing, the appropriate polymer solution into a mold set-up comprised of a mold and the reinforcing elements of the present invention. The mold set-up is cooled in an appropriate bath or on a refrigerated shelf and then lyophilized, thereby providing a reinforced tissue engineered scaffold. The biological component can be added either before or after the lyophilization step. In the course of forming the foam component, it is believed to be important to control the rate of freezing of the polymer-solvent system. The type of pore morphology that is developed during the freezing step is a function of factors such as the solution thermodynamics, freezing rate, temperature to which it is cooled, concentration of the solution, and whether homogeneous or heterogenous nucleation occurs. One of ordinary skill in the art can readily optimize the parameters without undue experimentation.

The required general processing steps include the selection of the appropriate materials from which the polymeric foam and the reinforcing components are made. If a mesh reinforcing material is used, the proper mesh density must be selected. Further, the reinforcing

material must be properly aligned in the mold, the polymer solution must be added at an appropriate rate and, preferably, into a mold that is tilted at an appropriate angle to avoid the formation of air bubbles, and the polymer solution must be lyophilized.

In embodiments that utilize a mesh reinforcing material, the reinforcing mesh has to be of a certain density. That is, the openings in the mesh material must be sufficiently small to render the construct sutureable or otherwise fastenable, but not so small as to impede proper bonding between the foam and the reinforcing mesh as the foam material and the open cells and cell walls thereof penetrate the mesh openings. Without proper bonding the integrity of the layered structure is compromised leaving the construct fragile and difficult to handle.

During the lyophilization of the reinforced foam, several parameters and procedures are important to produce implants with the desired integrity and mechanical properties. Preferably, the reinforcement material is substantially flat when placed in the mold. To ensure the proper degree of flatness, the reinforcement (e.g., mesh) is pressed flat using a heated press prior to its placement within the mold. Further, in the event that reinforcing structures are not isotropic it is desirable to indicate this anisotropy by marking the construct to indicate directionality. This can be accomplished by embedding one or more indicators, such as dyed markings or dyed threads, within the woven reinforcements. The direction or orientation of the indicator will indicate to a surgeon the dimension of the implant in which physical properties are superior.

As noted above, the manner in which the polymer solution is added to the mold prior to lyophilization helps contribute to the creation of a tissue implant with adequate mechanical integrity. Assuming that a mesh reinforcing material will be used, and that it will be positioned between two thin (e.g., 0.75 mm), shims it should be positioned in a substantially

flat orientation at a desired depth in the mold. The polymer solution is poured in a way that allows air bubbles to escape from between the layers of the foam component. Preferably, the mold is tilted at a desired angle and pouring is effected at a controlled rate to best prevent bubble formation. One of ordinary skill in the art will appreciate that a number of variables will control the tilt angle and pour rate. Generally, the mold should be tilted at an angle of greater than about 1 degree to avoid bubble formation. In addition, the rate of pouring should be slow enough to enable any air bubbles to escape from the mold, rather than to be trapped in the mold.

If a mesh material is used as the reinforcing component, the density of the mesh openings is an important factor in the formation of a resulting tissue implant with the desired mechanical properties. A low density, or open knitted mesh material, is preferred. One particularly preferred material is a 90/10 copolymer of PGA/PLA, sold under the tradename VICRYL (Ethicon, Inc., Somerville, NJ). One exemplary low density, open knitted mesh is Knitted VICRYL VKM-M, available from Ethicon, Inc., Somerville, NJ.

The density or "openness" of a mesh material can be evaluated using a digital photocamera interfaced with a computer. In one evaluation, the density of the mesh was determined using a Nikon SMZ-U Zoom with a Sony digital photocamera DKC-5000 interfaced with an IBM 300PL computer. Digital images of sections of each mesh magnified to 20x were manipulated using Image-Pro Plus 4.0 software in order to determine the mesh density. Once a digital image was captured by the software, the image was thresholded such that the area accounting for the empty spaces in the mesh could be subtracted from the total area of the image. The mesh density was taken to be the percentage of the remaining digital image. Implants with the most desirable mechanical properties were found to be those with a mesh density in the range of about 12 to 80 % and more preferably about 45 to 80%.

The biological component or effector of the issue repair stimulating implant can be incorporated within the implant before or after manufacture of the implant, or before or after the surgical placement of the implant.

Prior to surgical placement, the implant comprising a foam and reinforcement layer can be placed in a suitable container comprising the biological component. After an appropriate time and under suitable conditions, the implant will become impregnated with the biological component. Alternatively, the biological component can be incorporated within the implant by, for example, using an appropriately gauged syringe to inject the effectors into the implant. Other methods well known to those of ordinary skill in the art can be applied in order to load an implant with an appropriate biological component, such as mixing, pressing, spreading, and placing the biological component into the implant. Alternatively, the biological component can be mixed with a gel-like carrier prior to injection into the implant. The gel-like carrier can be a biological or synthetic hydrogels, incluging alginates, cross-linked alginates, hyaluronic acid, collagen gel, poly(N-isopropylacrylamide), poly(oxyalkylene), copolymers of poly(ethylene oxide)-poly(propylene oxide), and blends thereof.

Following surgical placement, an implant devoid of any biological component can be infused with effectors, or an implant with an existing biological component can be augmented with a supplemental quantity of the biological component. One method of incorporating a biological component within a surgically installed implant is by injection using an appropriately gauged syringe.

The amount of the biological component included with an implant will vary depending on a variety of factors, including the size of the implant, the material from which the implant is made, the porosity of the implant, the identity of the biologically component, and the intended purpose of the implant. One of ordinary skill in the art can readily determine the appropriate

quantity of biological component to include within an implant for a given application in order to facilitate and/or expedite the healing of tissue. The amount of biological component will, of course, vary depending upon the identity of the biological component and the given application.

FIGS. 4 and 5 illustrate a mold set up useful with the present invention in which mold 18 has a base 21 and side walls 22. Bottom shims 24 are disposed parallel to each other on an upper surface of base 21. Although parallel alignment of bottom shims 24 is illustrated, any number of shims, as well as any desired alignment, may be utilized. As further illustrated, reinforcing fabric 25 is placed over the bottom shims 24, and held in place by top shims 26, that are disposed parallel to each other on the reinforcing fabric 25. Though not shown, reinforcing fabric 25 can be placed between the bottom shims 24 and top shims 26 in a variety of ways. In one embodiment, the height of the bottom shims 24 can be varied so the mesh is placed nearer to the top or bottom surface of the sandwich construct.

In another embodiment, an electrostatically spun fabric barrier may be added to act as a barrier to hyperplasia and tissue adhesion, thus reducing the possibility of postsurgical adhesions. The fabric barrier is preferably in the form of dense fibrous fabric that is added to the implant. Preferably, the fibrous fabric is comprised of small diameter fibers that are fused to the top and/or bottom surface of the foam component. This enables certain surface properties of the structure, such as porosity, permeability, degradation rate and mechanical properties, to be controlled.

One of ordinary skill in the art will appreciate that the fibrous fabric can be produced via an electrostatic spinning process in which a fibrous layer can be built up on a lyophilized foam surface. This electrostatic spinning process may be conducted using a variety of fiber

materials. Exemplary fiber materials include aliphatic polyesters. A variety of solvents may be used as well, including those identified above that are useful to prepare the polymer solution that forms the foam component.

The composition, thickness, and porosity of the fibrous layer may be controlled to provide the desired mechanical and biological characteristics. For example, the bioabsorption rate of the fibrous layer may be selected to provide a longer or shorter bioabsorption profile as compared to the underlying foam layer. Additionally, the fibrous layer may provide greater structural integrity to the composite so that mechanical force may be applied to the fibrous side of the structure. In one embodiment the fibrous layer could allow the use of sutures, staples or various fixation devices to hold the composite in place. Generally, the fibrous layer has a thickness in the range of about 1 micron to 1000 microns. However, for some applications such as rotator cuff and meniscus injury repair, the fibrous layer has a thickness greater than about 1.5 mm.

In one embodiment of the present invention, the tissue repair stimulating implant is used in the treatment of a tissue injury, such as injury to a rotator cuff or meniscus. The implant can be of a size and shape such that it matches the geometry and dimensions of a desired portion or lesion of the tissue to be treated. As noted above, the biological component may be added to the implant during or after manufacture of the implant or before or after the implant is installed in a patent. An additional quantity of the biological component may be added after the implant is installed. Once access is made into the affected anatomical site (whether by minimally invasive or open surgical technique), the implant can be affixed to a desired position relative to the tissue injury, such as within a tear or lesion. Once the implant is placed in the desired position or lesion, it can be affixed by using a suitable technique. In one aspect, the implant can be affixed by a chemical and/or mechanical fastening technique.

Suitable chemical fasteners include glues and/or adhesive such as fibrin glue, fibrin clot, and other known biologically compatible adhesives. Suitable mechanical fasteners include sutures, staples, tissue tacks, suture anchors, darts, screws, and arrows. It is understood that combinations of one or more chemical and/or mechanical fasteners can be used. Alternatively, one need not use any chemical and/or mechanical fasteners. Instead, placement of the implant can be accomplished through an interference fit of the implant with an appropriate site in the tissue to be treated.

One of ordinary skill in the art will appreciate that the identity of the effector(s) that serve as the biological component may be determined by a surgeon, based on principles of medical science and the applicable treatment objectives.

The following examples are illustrative of the principles and practice of this invention. Numerous additional embodiments within the scope and spirit of the invention will become apparent to those skilled in the art.

Example 1

This example describes the preparation of three-dimensional elastomeric tissue implants with and without a reinforcement in the form of a biodegradable mesh.

A solution of the polymer to be lyophilized to form the foam component was prepared in a four step process. A 95/5 weight ratio solution of 1,4-dioxane/(40/60 PCL/PLA) was made and poured into a flask. The flask was placed in a water bath, stirring at 70°C for 5 hrs. The solution was filtered using an extraction thimble, extra coarse porosity, type ASTM 170-220 (EC) and stored in flasks.

Reinforcing mesh materials formed of a 90/10 copolymer of polyglycolic/polylactic acid (PGA/PLA) knitted (Code VKM-M) and woven (Code VWM-M), both sold under the tradename VICRYL were rendered flat by ironing, using a compression molder at 80 °C/2 min. Figure 6 is a scanning electron micrograph (SEM) of the knitted mesh. After preparing the meshes, 0.8-mm shims were placed at each end of a 15.3 x15.3 cm aluminum mold, and the mesh was sized (14.2 mm) to fit the mold. The mesh was then laid into the mold, covering both shims. A clamping block was then placed on the top of the mesh and the shim such that the block was clamped properly to ensure that the mesh had a uniform height in the mold. Another clamping block was then placed at the other end, slightly stretching the mesh to keep it even and flat.

As the polymer solution was added to the mold, the mold was tilted to about a 5 degree angle so that one of the non-clamping sides was higher than the other. Approximately 60 ml of the polymer solution was slowly transferred into the mold, ensuring that the solution was well dispersed in the mold. The mold was then placed on a shelf in a Virtis, Freeze Mobile G freeze dryer. The following freeze drying sequence was used: 1) 20°C for 15 minutes; 2) -5°C for 120 minutes; 3) -5°C for 90 minutes under vacuum 100 milliTorr; 4) 5°C for 90 minutes under vacuum 100 milliTorr; 5) 20°C for 90 minutes under vacuum 100 milliTorr. The mold assembly was then removed from the freezer and placed in a nitrogen box overnight. Following the completion of this process the resulting implant was carefully peeled out of the mold in the form of a foam/mesh sheet.

Nonreinforced foams were also fabricated. To obtain non-reinforced foams, however, the steps regarding the insertion of the mesh into the mold were not performed. The lyophilization steps above were followed.

Figure 7 is a scanning electron micrograph of a portion of an exemplary mesh-reinforced foam tissue implant formed by this process. The pores in this foam have been optimized for cell ingrowth.

Example 2

Lyophilized 40/60 polycaprolactone/polylactic acid, (PCL/PLA) foam, as well as the same foam reinforced with an embedded VICRYL knitted mesh, were fabricated as described in Example 1. These reinforced implants were tested for suture pull-out strength and burst strength and compared to both standard VICRYL mesh and non-reinforced foam prepared following the procedure of Example 1.

Specimens were tested both as fabricated, and after *in vitro* exposure. *In vitro* exposure was achieved by placing the implants in phosphate buffered saline (PBS) solutions held at 37°C in a temperature controlled waterbath.

For the suture pull-out strength test, the dimension of the specimens was approximately 5 cm X 9 cm. Specimens were tested for pull-out strength in the wale direction of the mesh (knitting machine axis). A size 0 polypropylene monofilament suture (Code 8834H), sold under the tradename PROLENE (by Ethicon, Inc., Somerville, NJ) was passed through the mesh 6.25 mm from the edge of the specimens. The ends of the suture were clamped into the upper jaw and the mesh or the reinforced foam was clamped into the lower jaw of an Instron model 4501. The Instron machine, with a 20lb load cell, was activated using a cross-head speed of 2.54 cm per minute. The ends of the suture were pulled at a constant rate until failure occurred. The peak load (lbs) experienced during the pulling was recorded.

The results of this test are shown below in Table 1.

Table 1: Suture Pull-Out Data (lbs)

Time	Foam	Mesh	Foamed Mesh
0 Day	0.46	5.3 +/- 0.8	5.7 +/-0.3
7 Day	-	4.0 +/-1.0	5.0 +/-0.5

For the burst strength test, the dimension of the specimens was approximately 15.25 cm X 15.25 cm. Specimens were tested on a Mullen tester (Model J, manufactured by B.F. Perkins, a Stendex company, a division of Roehlen Industries, Chicopee, MA). The test followed the standard operating procedure for a Mullen tester. Results are reported as pounds per square inch (psi) at failure.

The results of the burst strength test are shown in Table 2.

Table 2: Burst Strength Data (psi)

Time	Point-Knitted VICRYL Mesh	Foamed Knitted Mesh
0 Day	1349.5	1366.8
7 Day	1109.4	1279.6

Example 3

Mesh reinforced foam implants were implanted in an animal study and compared to currently used pelvic floor repair materials. The purpose of this animal study was to evaluate the subcutaneous tissue reaction and absorption of various polymer scaffolds. The tissue reaction and absorption was assessed grossly and histologically at 14 and 28 days post-implantation in the dorsal subcutis. In addition, the effect of these scaffolds on the bursting strength of incisional wounds in the abdominal musculature was determined. Burst testing

was done at 14 and 28 days on ventrally placed implants and the attached layer of abdominal muscle.

Lyophilized 40/60 polycaprolactone/polylactic acid (PCL/PLA) foam, as well as the same foam reinforced with an embedded VICRYL knitted mesh were fabricated as described in Example 1. The foam and mesh reinforced foam implant were packaged and sterilized with ethylene oxide gas following standard sterilization procedures. Controls for the study included: a VICRYL mesh control, a mechanical control (No mesh placed), and a processed porcine corium, sold under the tradename DermMatrix (by Advanced UroScience, St. Paul, MN) control.

The animals used in this study were female Long-Evans rats supplied by Harlan Sprague Dawley, Inc. (Indianapolis, Indiana) and Charles River Laboratories (Portage, Michigan). The animals weighed between 200 and 350 g. The rats were individually weighed and anesthetized with an intraperitoneal injection of a mixture of ketamine hydrochloride (sold under the tradename KETASET, manufactured for Aveco Co., Inc., Fort Dodge, Iowa, by Fort Dodge Laboratories, Inc., Fort Dodge, Iowa,) (dose of 60 milligram/kg animal weight) and xylazine hydrochloride (sold under the tradename XYLAZINE, Fermenta Animal Health Co., Kansas City, MO) (dose of 10 milligrams/kg animal weight). After induction of anesthesia, the entire abdomen (from the forelimbs to the hindlimbs) and dorsum (from the dorsal cervical area to the dorsal lumbosacral area) was clipped free of hair using electric animal clippers. The abdomen was then scrubbed with chlorhexidine diacetate, rinsed with alcohol, dried, and painted with an aqueous iodophor solution of 1% available iodine. The anesthetized and surgically prepared animal was transferred to the surgeon and placed in a supine position. Sterile drapes were applied to the prepared area using aseptic technique.

A ventral midline skin incision (approximately 3-4 cm) was made to expose the abdominal muscles. A 2.5 cm incision was made in the abdominal wall, approximately 1 cm caudal to the xyphoid. The incision was sutured with size 3-0 VICRYL suture in a simple continuous pattern. One of the test articles, cut to approximately 5 cm in diameter, was placed over the sutured incision and 4 corner tacks were sutured (size 5-0 PROLENE) to the abdominal wall at approximately 11:00, 1:00, 5:00 and 7:00 o'clock positions. The skin incision was closed with skin staples or metal wound clips.

After the surgeon completed the laparotomy closure, mesh implant, and abdominal skin closure, the rat was returned to the prep area and the dorsum was scrubbed, rinsed with alcohol, and wiped with iodine as described previously for the abdomen. Once the dorsum was prepped, the rat was returned to a surgeon and placed in the desired recumbent position for dorsal implantation. A transverse skin incision, approximately 2 cm in length, was made approximately 1 cm caudal to the caudal edge of the scapula. A pocket was made in the dorsal subcutis by separating the skin from the underlying connective tissue via transverse blunt dissection. One of the test materials cut to approximately 2.0 x 2.0 cm square, was then inserted into the pocket and the skin incision closed with skin staples or metal wound clips.

Each animal was observed daily after surgery to determine its health status on the basis of general attitude and appearance, food consumption, fecal and urinary excretion and presence of abnormal discharges.

The animals utilized in this study were handled and maintained in accordance with current requirements of the Animal Welfare Act. Compliance with the above Public Laws was accomplished by adhering to the Animal Welfare regulations (9 CFR) and conforming to the current standards promulgated in the Guide for the Care and Use of Laboratory Animals.

For the histopathology study, the rats were sacrificed after two weeks or four weeks, and the dorsal subcutaneous implant was removed, trimmed, and fixed in 10 % neutral buffered Formalin (20X the tissue volume). The samples were processed in paraffin, cut into 5 mm sections, and stained with Hematoxylin Eosin (H & E).

Dorsal samples for tissue reaction assessment were cut to approximate 2.0 cm squares. Ventral samples for burst testing were cut to approximate 5.0 cm diameter circles.

The bursting strength of each specimen was measured together with the attached underlying abdominal muscle layer following the method of Example 2. The results of the burst strength tests are shown in Table 3.

Table 3: Burst Strength (psi)

Sample	14 Days	28 Days
Mesh Reinforced Foam	81.8 +/-17.3	73 +/-4.5
Dermatrix	70 +/-4.0	70*

*Standard deviation is not available since only one sample survived until explant.

The histopathology study showed the mesh reinforced foam constructs had the highest degree of fibrous ingrowth and most robust encapsulation of all the implants tested at both time points. This fibrous reaction was mild in extent at 28 days.

Example 4

This example describes another embodiment of the present invention in which the preparation of a hybrid structure of a mesh reinforced foam is described.

A knitted VICRYL mesh reinforced foam of 60/40 PLA/PCL was prepared as described in Example 1. A sheet, 2.54 cm x 6.35 cm, was attached on a metal plate connected with a ground wire. The sheet was then covered with microfibrous bioabsorbable fabric produced by an electrostatic spinning process. The electrostatically spun fabric provides resistance to cell prevention from surrounding tissues and it enhances the sutureability of the implant.

A custom made electrostatic spinning machine located at Ethicon Inc (Somerville, NJ) was used for this experiment. A Spellman high voltage DC supply (Model No.: CZE30PN1000, Spellman High Voltage Electronics Corporation, Hauppauge, NY) was used as high voltage source. Applied voltage as driving force and the speed of mandrel were controlled. Distance between the spinneret and the plate was mechanically controlled.

A 14% solution of a 60/40 PLA/PCL copolymer produced at Corporate Biomaterials Center, a Division of Ethicon. Inc, Somerville, NJ was prepared in trichloroethane chloride (TEC) solvent. The polymer solution was placed into a spinneret and high voltage was applied to the polymer solution. This experiment was performed at ambient temperature and humidity. The operating conditions during spinning were as follows:

Spinneret voltage:	25,000 V
Plate voltage:	Grounded
Spinneret to mandrel distance:	15 cm

This process resulted in a deposited porous elastomeric polymer of approximately 10-500 μm in thickness on the surface of the mesh reinforced foam.

Example 5

Peel test specimens of mesh reinforced foam were made so as to separate otherwise bonded layers at one end to allow initial gripping required for a T-peel test (ref. ASTM D1876-95).

Copolymer foams of 40/60 polycaprolactone/polylactic acid (PCL/PLA), reinforced with both 90/10 copolymer of polyglycolic/polylactic acid (PGA/PLA) knitted (Code VKM-M) and woven (Code VWM-M) meshes, were fabricated as described in Example 1. Test specimens strips, 2.0 cm x 11.0 cm, were cut from the reinforced foam. Due to the cost of labor and materials, the size of the specimens was less than that cited in the above ASTM standard. The non-bonded section for gripping was produced by applying an aluminum foil blocker at one end to inhibit the penetration of polymer solution through the mesh reinforcement. The specimens were tested in an Instron Model 4501 Electromechanical Screw Test Machine. The initial distance between grips was 2.0 cm. The cross-head speed for all tests was held constant at 0.25 cm/min. The number of specimens of each construct tested was five.

The knitted VICRYL mesh foamed specimens required less force (0.087 +/- 0.050 in*lbf) to cause failure than did the woven VICRYL foamed specimens (0.269 +/- 0.054 in*lbf). It is important to note that the mode of failure in the two constructs was different. In the woven mesh specimens, there was some evidence of peel, whereas in the knitted mesh specimens, there was none. In fact, in the knitted specimens there was no sign of crack propagation at the interface between layers. A rate dependency in peel for the woven mesh specimens was noted. The test rate of 0.25 cm/min was chosen due to the absence of peel and swift tear of the foam at higher separation rates. Test results reported herein consist of tests run at this cross-head speed for both types of mesh. A slower speed of 0.025 cm/min was tried for the knitted mesh construct to investigate the possible onset of peel at sufficiently low

separation speeds. However, the slower speed did not result in any change in the mode of failure.

In conclusion, the higher density of the woven mesh inhibited extensive penetration of polymeric foam and resulted in the dissipation of energy through the peeling of the foam from the mesh when subjected to a T-peel test at a cross-head speed of 0.25 cm/min. In the case of the lower density knitted mesh construct, there appeared to be little to no separation of foam from the mesh. In these experiments it appeared that the load was wholly dissipated by the cohesive tearing of the foam.

Example 6

Primary chondrocytes were isolated from bovine shoulders as described by Buschmann, M.D. et al. (J.Orthop.Res.10, 745-752, 1992). Bovine chondrocytes were cultured in Dulbecco's modified eagles medium (DMEM-high glucose) supplemented with 10% fetal calf serum (FCS), 10 mM HEPES, 0.1 mM nonessential amino acids, 20 μg/ml L-proline, 50 μg/ml ascorbic acid, 100 U/ml penicillin, 100 μg/ml streptomycin and 0.25 μg/ml amphotericin B (growth media). Half of the medium was replenished every other day.

5 mm X 2mm discs or scaffolds were cut from reinforced foam polymer sheets (60/40 PLA/PCL foam reinforced with 90/10 PGA/PLA) prepared as described in Example 1. These discs were sterilized for 20 minutes in 70% ethanol followed by five rinses of phosphate-buffered saline (PBS).

Freshly isolated bovine chondrocytes were seeded at 5 x 10^6 cells (in 50 μl medium) by a static seeding method in hydrogel-coated plates (ultra low cluster dishes, Costar). Following 6 hours of incubation in a humidified incubator, the scaffolds were replenished with 2 ml of growth media. The scaffolds were cultured statically for up to 6 weeks in growth media.

Constructs harvested at various time points (3 and 6 weeks) were fixed in 10% buffered formalin, embedded in paraffin and sectioned. Sections were stained with Safranin-O (SO; sulfated glycosaminoglycans - GAG's) or immunostained for collagen type I and II. Three samples per time point were sectioned and stained.

Following 3-6 weeks of culturing under static conditions, the architecture of the scaffolds supported uniform cell seeding and matrix formation throughout the thickness of the scaffolds. Furthermore, the histological sections stained positively for Type II and GAG and weakly for collagen Type I indicating a cartilage-like matrix.

Example 7

Lyophilized 60/40 PLA/PCL foam, as well as the same foam reinforced with an embedded Vicryl (90/10 PGA/PLA) knitted mesh were fabricated analogous to the method described in Example 1, packaged and sterilized with ethylene oxide gas.

Animals were housed and cared for at Ethicon, Inc. (Somerville, NJ) under an approved institutional protocol. Three neutered male adult Neubian goats (50-65 Kg) were used in the study. An analgesic, Buprenorphine hydrochloride, was administered subcutaneously (0.005 mg/kg) about 2-3 hrs before the start of the surgery. Anesthesia was induced in each goat with an intravenous bolus of Ketamine at 11.0 mg/kg and Diazepam at 0.5 mg/kg both given simultaneously IV. Next, animals were intubated and maintained in a plane of deep anesthesia with 3% Isoflurane and an oxygen flow rate of 11-15 ml/kg/min. A gastric tube was placed to prevent bloating. Cefazolin sodium (20 mg/kg) was administered intravenously preoperatively.

A medial approach to the right stifle joint by osteotomy of the origin of the medial collateral ligament was taken to achieve full access to the medial meniscus. Approximately 60% of the central meniscus was excised in the red-white zone. The scaffold (+/- reinforced mesh) was secured in the defect (9 X 5 X 2mm) using 6 interrupted PROLENE sutures (6-0)

on a C-1 taper needle (Fig. 9). The joint capsule, fascial, and skin layers were closed with PROLENE-0 or VICRYL 2-0 sutures. Following the surgery, the goats were placed in a Schroeder-Thomas splint with an aluminum frame for 2 weeks to allow for partial weight bearing of the right stifle.

The animals were sacrificed after two weeks, and the medial meniscus was removed, trimmed, and fixed in 10 % neutral buffered Formalin (20X the tissue volume). The samples were processed in paraffin, cut into 5 μm sections, and stained with Hematoxylin Eosin (H & E).

At necropsy, all implants with the embedded knitted mesh structure remained intact, whereas those without any mesh did not remain intact or were completely lost from the defect site. Furthermore, the histological sections show evidence of tissue ingrowth at the interface between the reinforced scaffolds and the native meniscus. Due to partial or complete loss of the non-reinforced foams from the defect site there was little or no tissue ingrowth into the scaffolds.

One of ordinary skill in the art will appreciate further features and advantages of the invention based on the above-described embodiments. Accordingly, the invention is not to be limited by what has been particularly shown and described, except as indicated by the appended claims. All publications and references cited herein are expressly incorporated herein by reference in their entirety.

What is claimed is: